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## Comparative analysis of the microbial diversity in liquid and foaming layer in biogas reactors

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Foaming incidents have been recorded in many biogas plants causing severe operational, economical and environmental problems (Kougias et al., 2014). However, the foaming phenomenon in biogas reactors fed with agro-industrial wastes has not been extensively investigated, especially with respect to the microbial composition of the digesters (Moeller et al., 2012). In the cited literature, it has been reported that specific microorganisms, which are mainly filamentous (e.g. *Gordonia species*, *Microthrix parvicella*), are attached to biogas bubbles and transferred to the air/liquid interface of sludge reactors or wastewater treatment works (Ganidi et al., 2009). Once these microorganisms accumulate on the liquid surface, they initiate biosurfactants production due to their metabolic activity, leading to the decrease of the surface tension and thus generate foaming. The aim of the present study was to investigate the microbial diversity in the liquid versus the foaming layer in manure-based biogas reactors suffering by foaming incidents in order to elucidate potential role and contribution of the microorganisms in foam promotion.

The experimental work was carried out in three thermophilic continuous stirred tank reactors (CSTR) fed with manure and supplemental amounts of lipids, proteins and carbohydrates. Once foaming was formed in the reactors, samples from the liquid and foaming layer were obtained and screened using 16S rDNA sequencing.

The results of these analyses revealed that there are indeed some species that significantly vary their relative abundance in the foaming layer compared to the liquid one (e.g. *Methanoculleus sp.*, *Dialister sp.*). However, based on the cited literature and to the best of our knowledge there was not a direct correlation of these species with foaming. Further investigation is needed in order to define the properties of these species on foam generation. Finally, it was observed that particles of barley plant (that was contained in the raw manure as ingredient of animal nutrition) were accumulated in the foaming layer. It has been previously documented that barley contributes in stabilization of beer foam due to the activity of one of its proteins (Brey et al., 2003). For that reason, it could be hypothesised that the existence of barley particles in our reactors could contribute in foaming although their presence was also prior to the foaming incidents; their effect on foam formation or stabilization might be enhanced in correlation with other parameters (i.e. presence of specific microorganisms).

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